A National Survey to Determine Prevalence of *Trypanosoma cruzi* Infection among Pregnant Women in Ecuador


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Abstract. A nationwide survey was conducted to obtain an estimate of Chagas disease prevalence among pregnant women in Ecuador. As part of a national probability sample, 5,420 women seeking care for delivery or miscarriage at 15 healthcare facilities were recruited into the study. A small minority of participants reported knowing about Chagas disease or recognized the vector. A national seroprevalence of 0.1% (95% confidence interval [95% CI] = 0.0–0.2%) was found; cases were concentrated in the coastal region (seroprevalence = 0.2%; 95% CI = 0.0–0.4%). No cases of transmission to neonates were identified in the sample. Seropositive participants were referred to the National Chagas Program for evaluation and treatment. Additional studies are necessary to determine if areas of higher prevalence exist in well-known endemic provinces and guide the development of a national strategy for elimination of mother-to-child transmission of Chagas disease in Ecuador.

Chagas disease, caused by *Trypanosoma cruzi*, is spread mainly by hematophagous triatome vectors, blood transfusion, and congenital transmission. It is estimated that 200,000 people are infected with *T. cruzi* in Ecuador, where most provinces located on the Pacific coast as well Loja in the southern highlands have, for many decades, been known to be endemic for Chagas disease and continue to be active transmission foci. Meanwhile, sylvatic foci were detected in the northern Ecuadorian Amazon region during the 1990s, and additional studies have shown that this region is very active for *T. cruzi* transmission. However, despite punctual knowledge at the local scale, the global epidemiological situation of Chagas disease in Ecuador remains largely uncharacterized: no countrywide study exists to provide a global estimate of prevalence for the same sample and gain insight into the epidemiology of congenital transmission in the country. Five percent of *T. cruzi*-infected pregnant women transmit Chagas disease to their offspring congenitally, and over 15,000 such cases are estimated to occur yearly in Latin America. Screening of pregnant women in endemic areas for infection with *T. cruzi* is recommended. Furthermore, because of human migration, congenital Chagas transmission also constitutes a threat in the United States, Europe, and other non-endemic countries, and screening of migrant Latin American women in such settings has also been found to be cost-effective. However, parasitological treatment to infected women must be deferred until after pregnancy and breastfeeding, because nifurtimox and benznidazol (the only drugs available for treating Chagas disease) are potentially teratogenic and also, frequently cause undesirable secondary effects. Nonetheless, when newborns are treated before 1 year of age, infection is cleared in almost all cases without the secondary effects common among adults. Diagnosis in neonates is possible immediately after birth through parasitological methods, such as visualization of trypomastigotes on peripheral or cord blood, as well as blood concentration techniques (Strout or microhematocrit), whereas serologic diagnosis is possible starting at 8 months after birth, when potentially confounding maternal antibodies have disappeared. Additionally, recent data suggest that parasitological treatment of young infected women may help prevent congenital transmission later in life. During 2011 and 2012, under the coordination of the Pan-American Health Organization (PAHO) and the Ecuadorian Ministry of Health, a study was launched to obtain nationally representative estimates of human immunodeficiency virus (HIV) and syphilis prevalence among Ecuadorian pregnant women as well as estimates of coverage of preventive antenatal services. Several other national entities as well as academic institutions collaborated in the study. An opportunity thus arose to measure the prevalence of *T. cruzi* infection in the same sample and gain insight into the epidemiology of congenital Chagas disease transmission in Ecuador. A transversal descriptive study with a two-stage cluster sampling strategy and a target sample size of 6,000 women was designed and implemented (details are provided elsewhere). Briefly, healthcare facilities listed in the registry of deliveries and miscarriages of the Ecuadorian Ministry of Health were arranged according to region and province (to improve geographical representation), and smaller facilities (those reporting less than 400 births/miscarriages per year) were excluded. Using a systematic sampling with probability of selection proportional to the number of live births and miscarriages reported for each healthcare facility in 2008, 15 facilities were selected, including some within each of three major geographic regions of Ecuador: coastal region (8 facilities), highlands (6 facilities), and Amazon basin (1 facility). The number of participants recruited in each location was proportional to the number of pregnant women who received care at the study site (Figure 1). A design effect of 1.25 was considered because of the multistage sampling design. Other statistical analysis details are provided in the work by Sanchez-Gomez and others.
90% provided specimens for Chagas disease tests. In the survey, less than 10% of women reported having ever heard of Chagas disease, and among them, only one-half (50.1%) knew that it could be transmitted from mother to infant during pregnancy. Additionally, around 20% of women were able to recognize images of the vector, and 12.8% of women reported having seen it on their homes or peridomiciles.

A 1-mL serum aliquot from each participating woman was submitted to the national reference laboratory (Instituto Nacional de Salud Pública e Investigación [INSPI]; formerly Instituto Nacional de Higiene y Medicina Tropical Leopoldo Izquieta Pérez [INHMT]) in Guayaquil, where serological tests were performed according to national guidelines: an initial serological screening (recombinant enzyme-linked immunosorbent assay [Chagatest ELISA recombinante v3.0, WeinerLab, Rosario, Argentina]) was followed by a second serological test (immunoagglutination, Chagatest HAI, WeinerLab, Rosario, Argentina) performed on reactive samples. Seropositivity in both tests was considered diagnostic of *T. cruzi* infection. Discordant results were resolved with a third independent test by a different serological method (i.e., indirect immunofluorescence [IFA], Inmuno-Con Chagas, WAMA Diagnóstica, Sao Paulo, Brazil), where reactivity in two of three tests was considered diagnostic.

In total, 9 of 5,420 participating women were found to be seropositive for *T. cruzi* infection, corresponding to 0.1% prevalence (Table 2). All cases were found in the coastal region: eight cases in Guayas and one case in El Oro, two provinces that are well-known to be endemic for Chagas disease. Interestingly, among seropositive women, only two knew about Chagas disease before the study, and only one of them knew that it could be transmitted congenitally. Additionally, one other woman recognized the vector. None of them reported ever receiving blood transfusions, suggesting that they most likely were infected by other means. None of them answered positively to any other item of the Chagas disease survey.

Contact information provided by the seropositive women at their healthcare facilities was used by the National Chagas Program to either contact them by telephone or visit them at their domiciles for follow-up. Six seropositive women had provided valid contact information (five from Guayas and one from El Oro), and infection with *T. cruzi* was systematically studied for all of their offspring, including infants born during the study as well as all of their siblings (eight infants total). The sole exception was one woman from Guayas, whose neonate had died soon after delivery and who had no other offspring.

At the parasitology laboratories of the INHMT in Guayaquil, blood concentration techniques (microStrout) and serological tests (same techniques used for the mothers) were performed for all infants. Serology tests took place when the infants were at least 8 months of age. No evidence of presence of *T. cruzi* or
anti-*T. cruzi* antibodies was found. Mothers were referred to the National Chagas Program for evaluation and treatment.

This is the first study reporting prevalence of Chagas disease among a nationally representative sampling of pregnant women in Ecuador. Our data suggest a surprisingly low prevalence of infection by *T. cruzi* among women seeking medical attention for delivery or miscarriage in the Ecuadorian healthcare system. Consistent with our results, previous studies in which migrant Ecuadorian women were screened for Chagas disease in Spain did not report any cases. From among the provinces randomly included in our study, the concentration of seropositive cases in Guayas and El Oro is not surprising, because both of them have traditionally been known as endemic for Chagas. Additionally, given the small number of seropositive women encountered (9 of 5,420) and the low transmission rate of *T. cruzi* through the congenital route (5%), the absence of congenitally transmitted cases in our sample is not unexpected.

However, our results must be interpreted with caution, because some limitations related to our sampling strategy, which was originally designed to obtain HIV and syphilis estimates, could influence the estimated Chagas disease prevalence. For example, the study is not representative of women because some limitations related to our sampling strategy, our sample is not unexpected.

The low transmission rate of *T. cruzi* known as endemic for Chagas. Additionally, given the small concentration of seropositive cases in Guayas and El Oro is among the provinces randomly included in our study, the larger in rural areas. Conducted in 1987, indicated that this percentage is significantly larger in rural areas. The higher proportion of home births in rural areas and its potential impact over the lack of detection of congenital Chagas cases have been recently highlighted in Bolivia. However, smaller healthcare facilities (fewer than 400 deliveries and miscarriages/year), which are also common in rural areas, were excluded from the sampling frame because of logistic issues. Therefore, the rural population is underrepresented in our study. Chagas disease is known to be more prevalent in these settings, and therefore, the prevalence at national level could be underestimated.

Additional studies should target all relevant geographic areas where vectorial transmission occurs. These include provinces in the Ecuadorian Amazon region, such as Sucumbíos, Orellana, Pastaza, Morona-Santiago, and Zamora-Chinchipe, where the data available from the National Chagas Program/Ministry of Health as well as reports from academic groups show that Chagas disease is an emerging threat. Furthermore, they should include Loja province, a key active endemic region in the highlands. Generating epidemiological information from the high-risk areas should be a priority to inform the most cost-effective strategies aimed at eliminating mother-to-child transmission of Chagas disease in Ecuador. Finally, an education component designed to inform the population about Chagas disease and the risk of congenital transmission is required, which is highlighted by the results of our surveys.

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